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SUPPLEMENTARY EUROPEAN SEARCH REPORT

Application Number
EP 93 90 8732

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.5)
Y	WO-A-92 00091 (BIOLIGAND INC.) 9 January 1992 * the whole document * ---	1-42	C12Q1/70 C07K5/00 C07K13/00 G01N33/53
Y	WO-A-90 00626 (THE BAYLOR COLLEGE OF MEDICINE) 25 January 1990 * the whole document * ---	1-42	
Y	SCIENCE, vol.251, 15 February 1991 pages 767 - 778 S.P.A.FODOER ET AL. 'Light-Directed, Spatially Adressable Parallel Chemical Synthesis.' * the whole document * ---	1-42	
P,Y	WO-A-92 09300 (ITEREX PHARMACEUTICALS LTD. PARTNERSHIP) 11 June 1992 * abstract * ---	1-42	
E	WO-A-93 09668 (AFFYMAX TECHNOLOGIES N.V.) 27 May 1993 * the whole document * ---	1-42	TECHNICAL FIELDS SEARCHED (Int.Cl.5) C07K C12Q C07H C12N
E	WO-A-93 06121 (AFFYMAX TECHNOLOGIES N.V.) 1 April 1993 * the whole document * -----	1-42	

The supplementary search report has been drawn up for the claims attached hereto.

Place of search	Date of completion of the search	Examiner
THE HAGUE	20 July 1995	Scott, J

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
P : intermediate document

T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

& : member of the same patent family, corresponding document

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What Is Claimed Is:

1. A bifunctional molecule according to the formula A-B-C, wherein A is a chemical moiety, B is a linker molecule operatively linked to A and C, and C is an identifier oligonucleotide comprising a sequence of nucleotides which sequence identifies the structure of chemical moiety A.

2. The bifunctional molecule of claim 1 wherein A is a polymer comprising a linear series of chemical units represented by the formula $(X_n)_a$, wherein X is a single chemical unit in polymer A; and identifier oligonucleotide C is represented by the formula $(Z_n)_a$, wherein Z is a unit identifier nucleotide sequence within oligonucleotide C that identifies the chemical unit X at position n; and wherein

n is a position identifier for both X in polymer A and Z in oligonucleotide C having the value of 1+i where i is an integer from 0 to 10, such that when n is 1, X or Z is located most proximal to the linker, and a is an integer from 4 to 50.

3. The bifunctional molecule of claim 2 wherein said unit identifier nucleotide sequence Z has a length of from 2 to 8 nucleotides.

4. The bifunctional molecule of claim 1 wherein said chemical moiety A is an oligosaccharide, polypeptide, glycolipid, lipid, proteoglycan, glycopeptide or oligonucleotide.

5. The bifunctional molecule of claim 2 wherein said polymer A is a polypeptide, X is an amino acid residue in said polypeptide, and unit identifier nucleotide sequence Z is a hexanucleotide sequence that identifies the amino acid residue at position n in polypeptide A.

6. The bifunctional molecule of claim 5 wherein

said amino acid residue is selected from the group consisting of natural, modified and non-natural amino acids.

7. The bifunctional molecule of claim 2 wherein said identifier oligonucleotide C has a nucleotide sequence according to the formula P1-$(Z_n)_a$-P2, where P1 and P2 are nucleotide sequences that provide PCR primer binding sites adapted to amplify the polymer identifier oligonucleotide.

8. The bifunctional molecule of claim 7 wherein said P1 and P2 each contain a sequence that defines a restriction endonuclease site when present in a PCR-amplified duplex DNA fragment.

9. The bifunctional molecule of claim 8 wherein said restriction sites are located proximal to $(Z_n)_a$ relative to the PCR primer binding sites.

10. The bifunctional molecule of claim 9 wherein said restriction endonuclease sites form non-overlapping cohesive termini upon restriction endonuclease cleavage.

11. A library comprising a plurality of species of bifunctional molecules according to claim 1.

12. The library of claim 11 wherein said plurality of species is defined by the formula V^a, where V represents the number of different chemical units forming an alphabet of possible chemical units of X, and a is an exponent to V and represents the number of chemical units of X forming polymer A.

13. The library of claim 12 wherein X is a natural amino acid and V is 20.

14. The library of claim 12 wherein the number of chemical units (a) forming polymer A can be from 3 to 8.

15. The library of claim 12 wherein X is an amino acid and a is 6.

16. The library of claim 12 wherein X is an amino acid and said unit identifier nucleotide sequence Z has a length from 3 to 6 nucleotides.

17. The library of claim 11 wherein each of said species of bifunctional molecules in said plurality is present in molar equivalents of from 0.2 to 10.0.

18. The library of claim 12 wherein said identifier oligonucleotide C in each of said species of bifunctional molecules has a nucleotide sequence according to the formula $P1-(Z_n)_s-P2$, where P1 and P2 are nucleotide sequences that provide PCR primer binding sites adapted to amplify the identifier oligonucleotide, and where the nucleotide sequences of P1 and P2 are shared by all bifunctional molecule species in the library.

19. A method for identifying a chemical structure that participates in a preselected binding interaction with a biologically active molecule, said chemical structure being present in a library of bifunctional molecules according to claim 11, comprising the steps of:

a) admixing in solution said library of bifunctional molecules with the biologically active molecule under binding conditions for a time period sufficient to form a binding reaction complex;

b) isolating the complex formed in step (a); and

c) determining the nucleotide sequence of the identifier oligonucleotide in the isolated complex and thereby identifying the chemical structure that participated in the preselected binding interaction.

20. The method of claim 19 wherein said biologically active molecule is affixed to a solid support.

21. The method of claim 19 wherein said

biologically active molecule is operatively linked to a binding means capable of binding a binder molecule.

22. The method of claim 21 wherein said binding means is selected from the group consisting of biotin, protein A and magnetic beads.

23. The method of claim 19 wherein said determining comprises the steps of:

i) forming a polymerase chain reaction (PCR) amplification product from the sequence of the isolated identifier oligonucleotide; and

ii) determining the sequence of the PCR amplification product, thereby determining the sequence of the identifier oligonucleotide.

24. A method for preparing a library comprising a plurality of bifunctional molecules according to claim 11 comprising the steps of:

a) providing a linker molecule B having termini A' and C' according to the formula A'-B-C' that is adapted for reaction with a chemical precursor unit X' at termini A' and with a nucleotide precursor Z' at termini C';

b) conducting syntheses by adding chemical precursor unit X' to termini A' of said linker and adding precursor unit identifier oligonucleotide Z' to termini C' of said linker, to form a composition containing bifunctional molecules having the structure X_n-B-Z_n;

c) repeating step (b) on one or more aliquots of the composition to produce aliquots that contain a product containing a bifunctional molecule;

d) combining the aliquots produced in step (c) to form an admixture of bifunctional molecules, thereby forming said library.

25. The method of claim 24 wherein said steps (c) and (d) are repeated on the admixture of step (d)

to add an additional chemical unit X and corresponding identifier oligonucleotide Z to the bifunctional molecules in the admixture.

26. The method of claim 25 wherein said repetition of steps (c) and (d) are repeated on the admixture from 1 to 6 times, thereby forming a polymer A on said bifunctional molecules such that a is from 3 to 10.

27. The method of claim 19 wherein said linker molecule is a bifunctional solid support selected from the group consisting of bf-CPG or *o*-NB-bf-CPG.

28. The method of claim 24 wherein said linker molecule is a bifunctional solid support selected from the group consisting of bf-CPG or *o*-NB-bf-CPG.

29. A bifunctional solid support for synthesizing an oligopeptide/ oligonucleotide conjugate comprising:

a solid support, said solid support being of a type which is dispersible in aqueous solution,

a first linkage unit coupled to said solid support,

a second linkage unit coupled to said first linkage unit, and

a bifunctional unit coupled to said second linkage unit,

said bifunctional unit having a first leaving group employable for oligopeptide synthesis and a second leaving group employable for oligonucleotide synthesis,

said first leaving group being N-FMOC or its functional equivalent,

said second leaving group being O-DMT or its functional equivalent,

said second linkage unit being coupled to said first linkage unit by means of a bond cleavable

by exposure to concentrated aqueous ammonia,

said solid support, said first linkage unit, said second linkage unit, said cleavable bond, and said bifunctional unit, exclusive of said first and second leaving groups, each being substantially chemically unreactive to conditions employed by oligopeptide synthetic protocols using FMOC leaving groups and conditions employed by oligonucleotide synthetic protocols using O-DMT leaving groups.

30. A bifunctional solid support as described in claim 29 wherein said solid support is a controlled pore glass.

31. A bifunctional solid support as described in claim 29 wherein said bond between said first and said second linkage units is an alkyl ester.

32. A bifunctional solid support as described in claim 29 wherein said bifunctional unit being a serine residue having an amino end, a carboxyl end, and a hydroxyl end, said serine being coupled at its carboxyl end to said second linkage unit, being coupled at its amino end to said first leaving group, and being coupled at its hydroxyl end to said second leaving group.

33. A bifunctional solid support as described in claim 29 wherein:

said solid support is controlled pore glass,

said bond between said first and said second linkage units is an alkyl ester, and

said bifunctional unit being a serine residue having an amino end, a carboxyl end, and a hydroxyl end, said serine being coupled at its carboxyl end to said second linkage unit, being coupled at its amino end to said first leaving group, and being coupled at its hydroxyl end to said second leaving group.

34. A bifunctional solid support as described in claim 33 wherein:

said solid support is aminopropyl-CPG,

said first linkage unit includes a sarcosine linker coupled to the aminopropyl-CPG and a succinyl linker coupled to the sarcosine linker by means of an amide bond,

said second linkage unit includes an aminohexanol group coupled to said succinyl linker by means of an alkyl ester,

said bifunctional unit includes an L-serine residue, the amino end of the serine being coupled by means of an amide bond to said aminohexanol linker, the carboxyl end of said serine being coupled to an FMOC leaving group, the hydroxyl end of said serine being coupled to an O-DMT leaving group.

35. A bifunctional solid support as described in claim 29 further comprising:

a third linkage unit interposed between and coupled to said bifunctional unit and said first leaving group,

said third linkage unit being cleavable by exposure to ultra- violet light.

36. A bifunctional solid support as described in claim 35 wherein said third linkage unit includes a 3-nitro-4-O-ethyl benzoate group coupled by means of an amide bond to the amino end of said serine and coupled by means of an ester bond to an FMOC blocked amino acid.

37. A bifunctional solid support as described in claim 36 wherein:

said solid support is aminopropyl-CPG,

said first linkage unit includes a sarcosine linker coupled to the aminopropyl-CPG and a succinyl linker coupled to the sarcosine linker by means of an

amide bond,

said second linkage unit includes an aminohexanol group coupled to said succinyl linker by means of an alkyl ester, and

said bifunctional unit includes an L-serine residue, the amino end of the serine being coupled by means of an amide bond to said aminohexanol linker, the carboxyl end of said serine being coupled to an FMOC leaving group, the hydroxyl end of said serine being coupled to an O-DMT leaving group.

38. An element of a library of oligopeptide/oligonucleotide conjugates comprising:

a solid support, said solid support being of a type which is dispersible in aqueous solution,

a first linkage unit coupled to said solid support,

a second linkage unit coupled to said first linkage unit,

a bifunctional unit coupled to said second linkage unit,

an oligopeptide attached to said bifunctional unit, and

an oligonucleotide attached to said bifunctional unit.

39. An element of a library of oligopeptide/oligonucleotide conjugates as described in claim 38 further comprising a cleavable bond for coupling said first linkage unit to said second linkage unit, said cleavable bond being cleavable by exposure to concentrated aqueous ammonia.

40. An element of a library of oligopeptide/oligonucleotide conjugates as described in claim 39 further comprising a cleavable bond for coupling said bifunctional unit to said oligopeptide, said cleavable bond being cleavable by exposure to

ultraviolet light.

41. An element of a library of oligopeptide/oligonucleotide conjugates comprising:

a bifunctional unit,

an oligopeptide attached to said bifunctional unit, and

an oligonucleotide attached to said bifunctional unit.

42. An element of a library of oligopeptide/oligonucleotide conjugates as described in claim 41 further comprising a cleavable bond for coupling said bifunctional unit to said oligopeptide, said cleavable bond being cleavable by exposure to ultraviolet light.